






Research Article

Introducing *Muciflexus inthanonensis* gen. et sp. nov. and updates on *Ochronectria* (Hypocreales): New insights from leaf litter fungi in Doi Inthanon National Park, Northern Thailand

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Abstract

While exploring the leaf litter fungal diversity in Doi Inthanon National Park, Northern Thailand, we discovered a previously unknown lineage within Niessliaceae. *Muciflexus inthanonensis* **gen. et sp. nov.** is therefore introduced to accommodate this new lineage. The link between the sexual and asexual morphs of *Ochronectria thailandica* is also established, while the generic description of *Ochronectria* is updated. A polyphasic approach was employed, consisting of multi-locus phylogenetic analysis using ITS, LSU, *rpb2*, and *tef1-α* along with the morphological evidence to support the findings. *Muciflexus inthanonensis* resembles the spore morphology of *Phialoseptomonium* even though they are phylogenetically distant. *Muciflexus inthanonensis* forms a sister clade with *Trichonectria setadpressa*, characterised by a long branch, but differs in asexual morphology, possessing clusters of simple or branched conidiophores compared to the sporodochia-like structure of *T. setadpressa*. The discovery of the asexual morph of *Ochronectria thailandica* from a terrestrial habitat confirms the versatile nature of the fungus, which inhabits both aquatic and terrestrial environments. Here we establish the link between the sexual and asexual morphs of *Ochronectria* by revealing the holomorphic nature of *O. thailandica*. This study highlights the diversity of leaf litter fungi and the potential of discovering novel fungal species in northern Thailand.

Key words: Holomorphic fungi, Hypocreales, lignocellulolytic fungi, one new genus, saprobes

Introduction

Forest ecosystems are one of the most efficient ecosystems in terms of nutrient utilisation, exhibiting the fastest nutrient recycling through a combination of biological and chemical processes (Likens 2013; Niego et al. 2023). This enables forest ecosystems to meet the substantial nutrient demand

necessary for sustaining health and functioning (Du et al. 2024). Although some bacteria can decompose plant litter, fungi play a key role in decomposing and nutrient recycling in the lignocellulose matrix (Kjøller et al. 1982; Osono 2007; Wood et al. 2009). Forest trees produce a large amount of leaf litter rich in lignocellulosic compounds. Fungi decompose up to 90% of lignocellulose compounds (Osono 2007), efficiently releasing nutrients into a mobile state, where they become readily available for absorption (Bucher et al. 2004). Additionally, plants establish symbiotic relationships with nitrogen-fixing rhizobia as an adaptation to nutrient limitations (Hyde et al. 2018). However, leaf litter is one of the sources that returns the absorbed nutrients by trees back to the soil, and saprobic fungi play a crucial role in releasing the trapped nutrients back into the soil (Bucher et al. 2004). Thus, identifying the fungi involved in litter decomposition will give insights into nutrient cycling, assess ecosystem resilience, carbon sequestration, and soil fertility, and help to realise the global value of fungi (Hyde et al. 2018).

Hyde and coworkers have been exploring the diversity of micro- and macro-fungi in northern Thailand for almost two decades; more than 500 fungi have been reported from this region by 2018 (Hyde et al. 2018). However, the continued exploration and discoveries have resulted in the number increasing since then (Thitla et al. 2022; Silva et al. 2023; Louangphan et al. 2024). The novelty of microfungi is not as high as that of macrofungi due to less exploration (Hyde et al. 2018). This makes northern Thailand a promising location for exploring microfungi, as many more species are yet to be discovered (Hyde et al. 2024).

Doi Inthanon National Park is a conserved area in northern Thailand representing a mountainous region of Chiang Mai Province (Teejuntuk et al. 2003). The changing geographical features and altitudes create diverse microenvironments that support various organisms and their associated diversity, including microfungi. This makes it a promising location for sheltering diverse species with different micro requirements. Identifying new fungal taxa is important as it expands the horizons of available possibilities and provides more opportunities in fields such as medicine, biochemistry, agriculture, and the function of natural phenomena in the environment (Hyde et al. 2018, 2019, 2024). Identifying novel lineages is crucial for advancing existing taxonomic knowledge and resolving uncertain taxonomic placements. Hence, discovering the missing lineages might serve as a fundamental approach to resolving such taxa. Exploring less-studied environments holds immense potential for discovering previously unknown species. Within the Hypocreales, despite being a well-established taxon, ongoing research continues to reveal new lineages, underscoring the unexplored biodiversity yet to be discovered (Hou et al. 2023; Perera et al. 2023; Sun et al. 2023).

Hypocreales can be found globally in various biotrophic, hemibiotrophic, saprobic, or hypersaprobic habitats (Lombard et al. 2015; Perera et al. 2023). This order is recognised for hosting many fungi that are important in agriculture and medicine (Rossman 1996; Lombard et al. 2015). According to the current update, 29 families are accepted in Hypocreales (Hyde et al. 2024). The members of Hypocreales exhibit a remarkable diversity in lifestyles and habitats (Perera et al. 2023). Hypocreales are generally composed of members with perithecial ascomata, while some genera are

cleistothecial (Rossman et al. 1999; Perera et al. 2023). Hypocreales are characterised by transparent, white, pale, bright, or darkly coloured, KOH \pm , LA \pm , soft, fleshy, or tough ascomata. They can be found superficially on the substrate or embedded within it, sometimes positioned in a stroma that ranges from weak to well-developed. Asci are unitunicate with 2–8 spores. The ascospores range from aseptate to having multiple septa, and they can sometimes be muriform. These ascospores can remain whole or disarticulate. The asexual morph of Hypocreales is usually hyphomycetous, rarely ceolomycetous (Rossman et al. 1999; Hyde et al. 2020; Perera et al. 2023).

During our ongoing exploration of fungal diversity in Doi Inthanon National Park, a comprehensive study on leaf litter led us to discover a previously undocumented genus-level lineage along with the asexual morph of *Ochronectria thailandica*. Here we amend the *Ochronectria* description by including the asexual morph characteristics with the available sexual morph characteristics. This paper provides a detailed account of their morpho-molecular characteristics while presenting the novelty of these findings. These findings provide insights into the lignicolous fungal community in northern Thailand and establish the holomorphic nature of the *Ochronectria thailandica*.

Materials and methods

Collection, morphological observation, and isolation

Leaf litter that had fallen onto the ground was collected from Doi Inthanon National Park, located in the Chiang Mai District of northern Thailand. The collection information was recorded (Rathnayaka et al. 2024), and the samples were taken to the laboratory at the Centre of Excellence in Fungal Research in paper bags. As detailed by Chomnunti et al. (2014), the leaf samples were incubated to stimulate additional sporulation. Subsequently, a morphological examination was conducted, followed by the isolation of single spores using a spore suspension technique (Senanayake et al. 2020). Spores were isolated into Potato Dextrose Agar (PDA) and Malt Extract Agar (MEA) media plates. The morphological examination of host samples was conducted using a Leica eZ4 educational stereo microscope (Leica, Wetzlar, Germany) and a Nikon ECLIPSE Ni compound microscope (Nikon, Tokyo, Japan). Images were captured using the Nikon dS-ri2 digital camera. Measurements were done using the Tarosoft (R) Image Frame Work Program V.09. Adobe Photoshop CS6 Extended software (Adobe Systems, USA) was used to process and present the resulting images. Herbarium materials and live cultures were deposited at the Mae Fah Luang University Herbarium (MFLU) and Mae Fah Luang University Culture Collection (MFLUCC), respectively.

DNA extraction and polymerase chain reaction (PCR) amplification

DNA was extracted using the E.Z.N.A.® tissue DNA Kit. The manufacturer's instructions were followed. Young cultures were used when they were around 1–2 months old. Approximately 30 mg of mycelia was used as the starting material for the DNA extraction.

Four loci, internal transcribed spacer regions (ITS), large subunit rRNA gene (LSU), DNA-directed RNA polymerase II subunit two gene (*rpb2*), and translation elongation factor 1-alpha gene (*tef-1α*) were amplified using PCR. The primers used and PCR conditions are listed in Table 1. Amplification was performed in a total reaction volume of 25 µl, consisting of 1 µl of genomic DNA template, 1 µl of each forward and reverse primer at a concentration of 20 µm, and 9.5 µl of double-distilled, deionised water, with 12.5 µl of 2× GoTaq® Green Master Mix (PROMEGA, USA). Sequencing was performed by the SolGent Co., Ltd. (South Korea).

Phylogenetic analyses

Forward and reverse sequences were assembled using the Staden Package (Staden et al. 2003), and the resulting sequences were compared against the NCBI GenBank database (Sayers et al. 2020). Related reference sequences were downloaded from the GenBank database (Suppl. material 1). Individual data sets referring to each gene region were aligned using MAFFT version 7 with the --auto flag (Kuraku et al. 2013; Katoh et al. 2019). The sequences were trimmed automatically using trimAl 1.2rev57 with the -gt (0.5) option (Capella-Gutiérrez et al. 2009). Sequence Matrix was used to concatenate the alignments in the order of ITS, LSU, *rpb2*, and *tef1-α* (Vaidya et al. 2011). The best-fit model for each individual dataset was selected using jModelTest2 (Guindon and Gascuel 2003; Darriba et al. 2012). Concatenated data sets were analysed using Maximum Likelihood (ML) and Bayesian Inference (BI) analyses.

The Maximum Likelihood tree was generated using the IQ-Tree web server, available at <http://iqtree.cibiv.univie.ac.at/> (Trifinopoulos et al. 2016). Bayesian inference was performed on the CIPRES Science Gateway platform using the MrBayes 3.2 tool (Huelsenbeck and Ronquist 2001; Ronquist and Huelsenbeck 2003; Ronquist et al. 2012). It was performed by four simultaneous Markov Chain Monte Carlo (MCMC) chains in two runs. Each run consisted of ten million generations, and trees were sampled every 1,000 generations. The first and final 25% results were discarded as the burn-in. The rest was used to calculate the BI posterior probabilities (PP). The final consensus phylograms were visualised using the FigTree drawing tool (version 1.4.0, Rambaut 2012) and edited using Microsoft PowerPoint. The guidelines of Maharachchikumbura et al. 2021 were used for introducing new taxa.

Pairwise evolutionary divergence between sequences was estimated using MEGA 12 (Kumar et al. 2024). Analyses were conducted using the Kimura 2-parameter model, with rate variation among sites modelled using a gamma distribution (shape parameter = 1). The partial deletion option was applied to eliminate all positions with less than 95% site coverage (Kimura 1980).

Results

***Muciflexus* V. S. Silva & Jayaward., gen. nov.**

Index Fungorum: IF903024

Facesoffungi Number: FoF17005

Classification. Niessliaceae, Hypocreales, Hypocreomycetidae, Sordariomycetes.

Etymology. "Muci-" refers to the slimy nature of the conidial masses, and "flexus" refers to the flexibility or branching of the conidiophores.

Table 1. Loci, primers, and PCR conditions used in this study.

Loci	PCR Primers	Sequence (5'–3')	PCR Cycles	References
ITS	ITS5	GGA AGT AAA AGT CGT AAC AAG G	(95 °C: 30 s, 55 °C: 50 s, 72 °C: 1 min) × 35 cycles	White et al. (1990)
	ITS4	TCC TCC GCT TAT TGA TAT GC		
LSU	LR0R	GTA CCC GCT GAA CTT AAG C	(95 °C: 30 s, 52 °C: 30 s, 72 °C: 1 min) × 35 cycles	Rehner and Samuels (1994); Vilgalys and Hester (1990)
	LR5	TCC TGA GGG AAA CTT CG		
<i>tef1</i> – <i>α</i>	EF1–983F	GCY CCY GGH CAY CGT GAY TTY AT	(95 °C: 30 s, 55 °C: 50 s, 72 °C: 1 min) × 35 cycles	Rehner and Buckley (2005)
	EF1–2218R	AT GAC ACC RAC RGC RAC RGT YTG		
<i>rpb2</i>	RPB25F2	GGG GWG AYC AGA AGA AGGC	(95 °C: 1 min, 52 °C: 30 s, 72 °C: 2 min) × 35 cycles	Sung et al. (2007)
	RPB27CR	CCC ATR GCT TGY TTR CCC AT		

Description. Surface mycelium composed of hyaline, smooth-walled, branched hyphae. Conidiophores arising directly from hyphae, straight to flexuous, erect, branched, hyaline, arranged in dense clusters or solitarily, cylindrical and slightly tapering towards the apical end. Conidiogenous cells integrated, adhering in slimy masses, apically produce conidia. Conidia often aseptate or multiseptate, granular, fusoid, apex obtuse, base truncate, straight to slightly curved, hyaline, smooth-walled.

Type species. *Muciflexus inthanonensis* V. S. Silva, K.D. Hyde & Jayaward.

***Muciflexus inthanonensis* V. S. Silva, K.D. Hyde & Jayaward., sp. nov.**

Index Fungorum: IF903018

Facesoffungi Number: FoF17006

Fig. 1

Holotype. MFLU 24–0382.

Etymology. “inthanonensis” refers to the type locality, Doi Inthanon, Thailand.

Description. Saprobic on a leaf of a fallen unknown broadleaf species. Sexual morph: Undetermined. Asexual morph: Hyphomycetous, erect, appear in solitary or in groups. Surface mycelium composed of branched, hyaline, smooth-walled hyphae. Conidiophores erect, simple or occasionally branched, straight to flexuous, arranged in dense clusters or solitary, arising directly from hyphae, with 0–1 basal septa, cylindrical, slightly tapering towards the apical end, hyaline, smooth-walled, 19.5–85 (\bar{x} = 55.3, n = 10) μm , base 2.2–3.2 (\bar{x} = 2.6, n = 10) μm , apex 1.3–1.9 (\bar{x} = 1.5, n = 10) μm . Conidiogenous cells integrated, apically produce 1–4 conidia, adhering in slimy masses. Conidia solitary or occasionally grouped, straight to slightly curved, often aseptate or 1-septate or rarely 2–3 septate, granular, fusoid, apex obtuse, base truncate, hyaline, smooth-walled, 10.5–19 (\bar{x} = 13.9, n = 68) \times 2.1–3.3 (\bar{x} = 2.7, n = 68) μm , L/W 5.14.

Culture characteristics. In both PDA and MEA media, culture diameter reaches an average of 6 mm within 5 days. On both media, colonies are flat. After about 45 days on the PDA media in the front, it develops in the buff with sparse white aerial mycelium at the centre. On the reverse also, it develops into buff with a smooth, entire margin. On the MEA media in front view, it develops into sparse mycelium white at the centre with an orangish margin ring extending to white aerial mycelium. On the reverse, it is buff and concentric rings gradually becoming light.

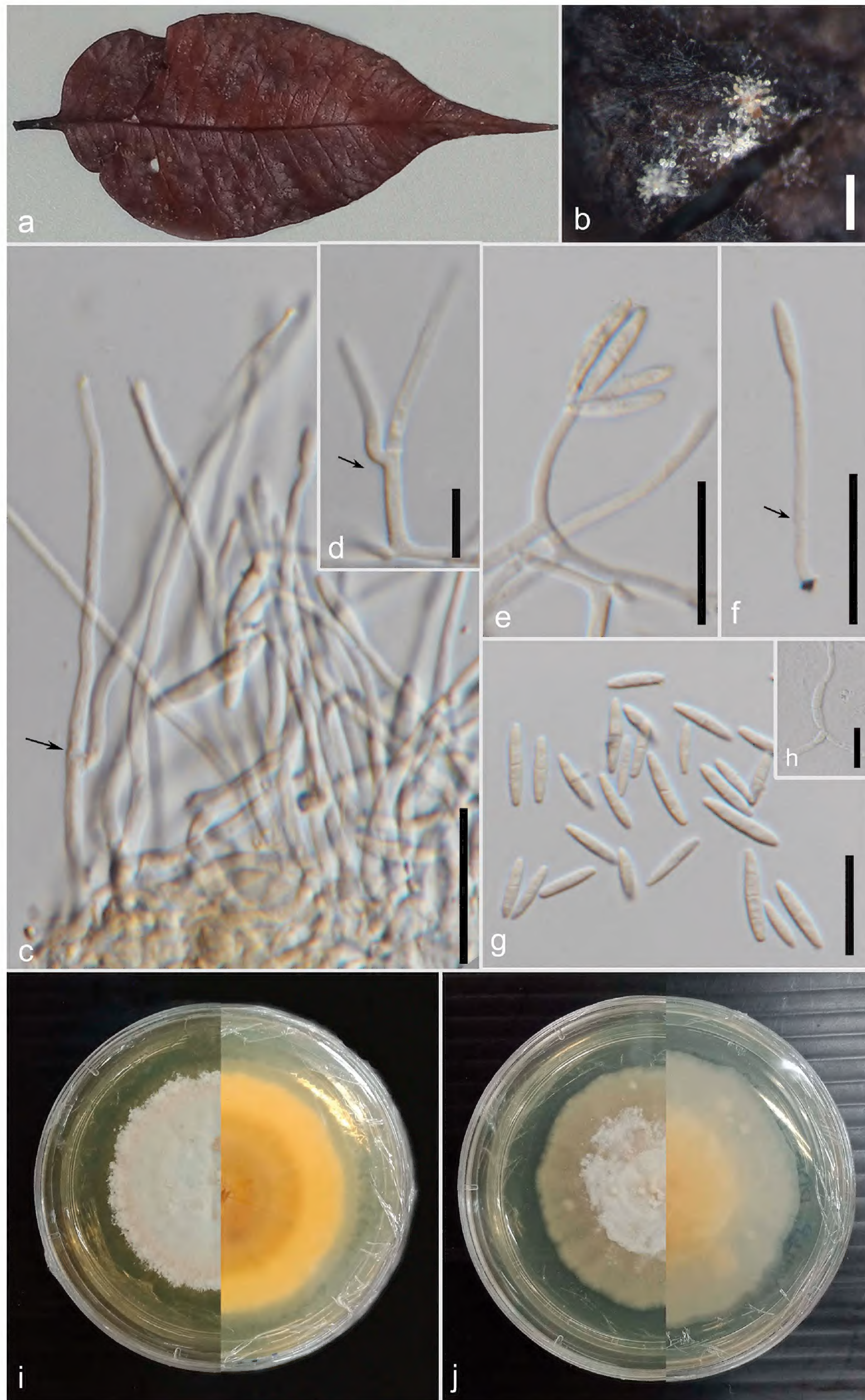


Figure 1. *Muciflexus inthanonensis* (Holotype MFLU24–0382) **a** host **b** a closer view of the colonies on the host substrate **c–f** conidiophores with attached conidia (arrows **c–d** branched conidiophore, **f** basal septation of conidiophore) **g** conidia **h** germinated spore **i** front and the reverse view of the culture on MEA media **j** front and the reverse view of the culture on PDA media. Scale bars: 200 μm (**b**); 20 μm (**c**, **e**, **f**); 10 μm (**d**, **g**, **h**).

Material examined. THAILAND • Chiang Mai Province, Doi Inthanon National Park, on a fallen unidentified broadleaf species, 30 November 2022, V. S. Hittanadurage Silva, V046 (holotype MFLU 24–0382); ex-type living culture (MFUL-CC 24–0502).

GenBank accession numbers. ITS: PQ528132, LSU: PQ528133, SSU: PQ528134, *rpb2*: PQ590309, *tef1*–*α*: PQ568247.

In the phylogenetic tree, the dataset comprised 58 strains representing Niessliaceae, including *incertae sedis* taxa. Following Hou et al. (2023), taxa with the species name, which are not yet formally accepted or validated under the International Code of Nomenclature for fungi, were also included. They are represented within “”. The outgroup is represented by four taxa from Nothoacremoniaceae (CBS 416.68, CBS 190.70, CBS 587.73, and CBS 397.70). The final concatenated nucleotide alignment was composed of ITS, LSU, *rpb2*, and *tef1*–*α* with 2974 sites in total (ITS = 1–569; LSU = 570–1410; *rpb2* = 1411–2166; *tef1*–*α* = 2167–2974). The maximum likelihood and Bayesian analyses yielded similar tree topologies, which are combined in Fig. 2. The maximum likelihood tree default setting in the IQ-TREE web server was used, and for the BI tree, the combined region run quality was checked using Tracer v1.7.2 after the runs were completed. All runs were conducted with effective sample size (ESS) values for all parameters. The alignment contained 1,340 unique sites (ITS = 357; LSU = 273; *rpb2* = 406; *tef1*–*α* = 304).

Notes. Hou et al. (2023) conducted phylogenetic analyses identifying two distinct clades (Clade G & Clade H) within the family Niessliaceae. The authors highlighted that the phylogenetic relationships within Niessliaceae remain unresolved, with several genera, viz., *Niesslia*, *Cylindromonium*, and *Trichonectria*, exhibiting polyphyletic characteristics (Hou et al. 2023).

In our analysis, *Muciflexus inthanonensis* clustered within H. Niessliaceae (Hou et al. 2023), alongside taxa from *Cylindromonium*, *Trichonectria*, and *Phialoseptomonium*. However, *Trichonectria* is currently classified as Hypocreales genera *incertae sedis* (Perera et al. 2023; Hyde et al. 2024). *Muciflexus inthanonensis* formed a sister clade to *Trichonectria setadpressa*, with a BYPP of 0.98 and MLBS of 89% statistical support. The noticeable branch length difference suggests that *M. inthanonensis* forms a distinct lineage, which may be attributed to the inclusion of two additional gene regions (*rpb2* and *tef1*–*α*) not available for *T. setadpressa* and potential genetic novelty. Morphologically, *T. setadpressa* is characterised by sporodochia-like conidiomata with subglobose to broadly ellipsoidal conidia as its asexual morph (Flakus et al. 2019), whereas *M. inthanonensis* produces clusters of simple or occasionally branched conidiophores with fusoid conidia. Additionally, *Trichonectria* is a lichenicolous genus (Perera et al. 2023), while *M. inthanonensis* is saprobic, found on a fallen unidentified broadleaf species. Furthermore, a pairwise comparison of all gene regions between *M. inthanonensis* and *T. setadpressa* (Table 2) provides further evidence supporting the genetic distinctiveness of the newly introduced genus.

Morphologically, *Muciflexus inthanonensis* closely resembles *Phialoseptomonium*, particularly in spore characteristics. Its solitary fusoid conidia, which are granular, hyaline, smooth-walled, and adhere in slimy masses, are similar to those of *Phialoseptomonium* (Crous et al. 2019a). However, *M. inthanonensis* can be distinguished by its aseptate or 1–3-septate, grouped conidia and comparatively smaller spore size (*P. eucalypti*: L/W = 6.7, *P. junci*: L/W = 6.2,

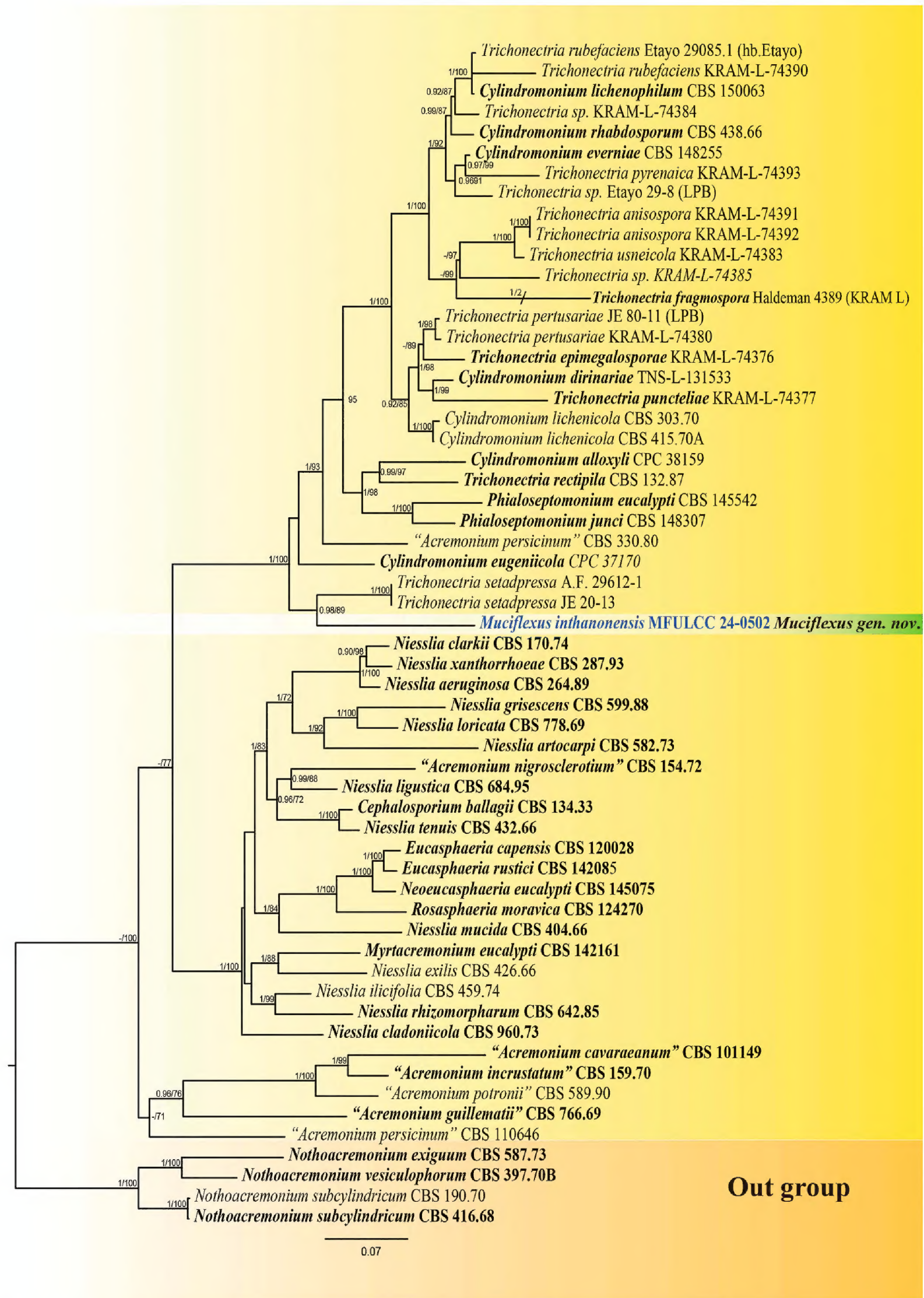


Figure 2. Maximum likelihood phylogenetic tree generated from ITS, LSU, *rpb2*, and *tef1*– α sequence data of selected taxa from Niessliaceae. The tree is rooted with four taxa from Nothoacremoniaceae (CBS 416.68, CBS 190.70, CBS 587.73, and CBS 397.70B). The newly generated sequence is in bold blue. Holotype and ex-type strains are in bold text. Bayesian posterior probabilities (BYPP) ≥ 0.95 and maximum likelihood bootstrap (MLBS) values $\geq 70\%$ are shown at the nodes.

M. inthanonensis: L/W = 5.14). Phylogenetically, *M. inthanonensis* clusters distantly from *Phialoseptomonium*. While *Phialoseptomonium* species have been reported as saprobes, taxonomic differentiation based on genetic data is also crucial. According to Raja et al. 2017, variation in the LSU gene region is

Table 2. Pairwise base pair (Bp) comparison of *Muciflexus inthanonensis* to other related taxa.

	<i>Cylindromonium eugeniicola</i> (CPC 37170)		<i>Trichonectria setadpressa</i> (JE 20–13)		<i>Phialoseptomonium eucalypti</i> (CBS 145542)		<i>Phialoseptomonium junci</i> (CBS 148307)	
	Bp differences	Gaps	Bp differences	Gaps	Bp differences	Gaps	Bp differences	Gaps
ITS	96/588(16%)	31/588(5%)	95/573(17%)	36/573(6%)	104/588(18%)	36/588(6%)	99/588(17%)	38/588(6%)
LSU	10/414(2%)	0/414(0%)	7/402(2%)	2/402(0%)	15/424(4%)	0/424(0%)	20/408(5%)	3/408(0%)
<i>tef1</i> – <i>α</i>	66/768(9%)	2/768(0%)	N/A	N/A	81/770(11%)	6/770(0%)	N/A	N/A
<i>rpb2</i>	N/A	N/A	N/A	N/A	No significant similarity		N/A	N/A

N/A – Sequence is not available.

indicative of differences at intermediate taxonomic levels, such as family and genus. Pairwise comparisons between *M. inthanonensis* and *Phialoseptomonium* species (Table 2) further support the likelihood that *M. inthanonensis* does not belong to the same genus.

Cylindromonium exhibits polyphyletic behaviour and was established to accommodate *Acremonium*-like taxa characterised by unbranched, hyaline conidiophores and cylindrical conidia (Crous et al. 2019b). Apart from *C. alloxyl*i and *C. eugeniicola*, the remaining members of the genus are lichenicolous (Suppl. material 2). *Cylindromonium alloxyl*i is mycophilic on *Meliola* and was found on *Alloxylon pinnatum* leaves (Crous et al. 2020), whereas *C. eugeniicola* is saprobic on leaves (Crous et al. 2019). Crous et al. 2019b mentioned the morphological resemblance between *Cylindromonium* and *Phialoseptomonium*; however, they can be distinguished by the cylindrical conidia of *Cylindromonium*. Based on this, pairwise genetic distances were calculated among all *Cylindromonium* and *Phialoseptomonium* species (Suppl. material 3), with the resulting *p*-distance values presented in Table 3. These values were then compared against the *p*-distance values of *Muciflexus inthanonensis* with both *Cylindromonium* and *Phialoseptomonium*, yielding the following results: ITS: 0.14–0.19, LSU: 0.12–0.06, *tef1*–*α*: 0.12–0.15, and *rpb2*: 0.31–0.40. These values fall within or exceed the range that differentiates *Cylindromonium* and *Phialoseptomonium*. Additionally, *Muciflexus* can be morphologically distinguished from *Cylindromonium* by its occasionally branched conidiophores and fusoid, occasionally grouped conidia. These combined morphological and phylogenetic differences provide strong support for the novelty of the proposed genus.

Based on these host associations along with morphological and phylogenetic evidence, it is inconclusive to place *Muciflexus inthanonensis* in any of the genera within the H clade of Niessliaceae (Hou et al. 2023). Therefore, here we propose a new genus, *Muciflexus*, to accommodate *Muciflexus inthanonensis*.

Table 3. Range of *p*-distance values of separating *Cylindromonium* from *Phialoseptomonium*.

	p-distance value	
	Maximum	Minimum
ITS	0.14	0.09
LSU	0.05	0.02
<i>tef1</i> – <i>α</i>	0.16	0.09
<i>rpb2</i>	0.33	0.26

***Ochronectria* Rossman & Samuels, emend V. S. Silva & Jayaward.**

Index Fungorum: IF28315

Facesoffungi Number: FoF13003

Classification. Bionectriaceae, Hypocreales, Hypocreomycetidae, Sordariomycetes.

Remarks. *Ochronectria* was established by Rossman et al. (1999), accommodating *Ochronectria calami* as the type species. The genus features subglobose to globose ascomata that become cupulate upon drying, peridium with three distinct layers, clavate asci, and fusiform ascospores containing guttules (Rossman et al. 1999; Lechat 2010; Li et al. 2016). Three species are accepted under *Ochronectria*, with no report on the asexual morph (Index Fungorum 2025). Discovery of the asexual form of *Ochronectria thailandica* in this study reveals the asexual morph of *Ochronectria*. Therefore, the genus description is emended here with general asexual morphology.

Description. Sexual morph: as described by the original description, Rossman et al. (1999)

Asexual morph: Hyphomycetous. Colonies on the host are solitary to gregarious, and the vegetative mycelium is superficial. Conidiophores erect, mononemotous, unbranched, 2-septate, hyaline, smooth-walled. Conidiogenous cells elongate, hyaline, holoblastic, and apically produce monoblastic conidia. Conidia aseptate, ellipsoidal to cylindrical, rarely ovoid, hyaline, smooth-walled.

***Ochronectria thailandica* Q.J. Shang & K.D. Hyde**

Index Fungorum: IF551918

Facesoffungi Number: FoF01815

Fig. 3

Description. Sexual morph: see Li et al. (2016). **Asexual morph:** saprobic and hyphomycetous. Colonies on the host solitary to gregarious, vegetative mycelium superficial. Conidiophores erect, mononemotous, smooth, hyaline, unbranched, 2-septate, 70–89 (\bar{x} = 77, n = 9) μ m. Conidiogenous cell elongated, hyaline, holoblastic, apically producing monoblastic conidia, 22–37 (\bar{x} = 28, n = 9) μ m. Conidia hyaline, smooth-walled, aseptate, ellipsoidal to cylindrical rarely ovoid, 4.5–8.5 \times 2–3 (\bar{x} = 6–2.5, n = 40) μ m, L/W 2.5.

Culture characteristics. Conidia germinating on MEA within 24 hours. Colonies on MEA reaching 1.4 mm diam. within 6 days in the dark at 25 °C, edge entire, flat or effuse, sparse. After 7 days colonies become white on the front face, and from below, reddish yellow gradually becomes slightly dark when mature.

Material examined. THAILAND • Chiang Mai Province, Doi Inthanon National Park, on a petiole of a fallen broadleaf species, 30 November 2022, Veenavee Silva, V054a, MFLU 24–0383, MFULCC 24–0503

GenBank accession numbers. ITS: PQ454717, LSU: PQ454721

In the phylogenetic analysis, the dataset consisted of 36 strains from Bionectriaceae, with two taxa from Stromatonectriaceae (CBS 125579 and CBS 127387) serving as the outgroup. The final concatenated nucleotide alignment included ITS, LSU, *rpb2*, and *tef1-a* sequences, totalling 2854 sites (ITS: 1–514; LSU: 515–1290; *rpb2*: 1291–2046; *tef1-a*: 2047–2854). Both maximum likelihood and Bayesian analyses produced similar tree topologies; they were

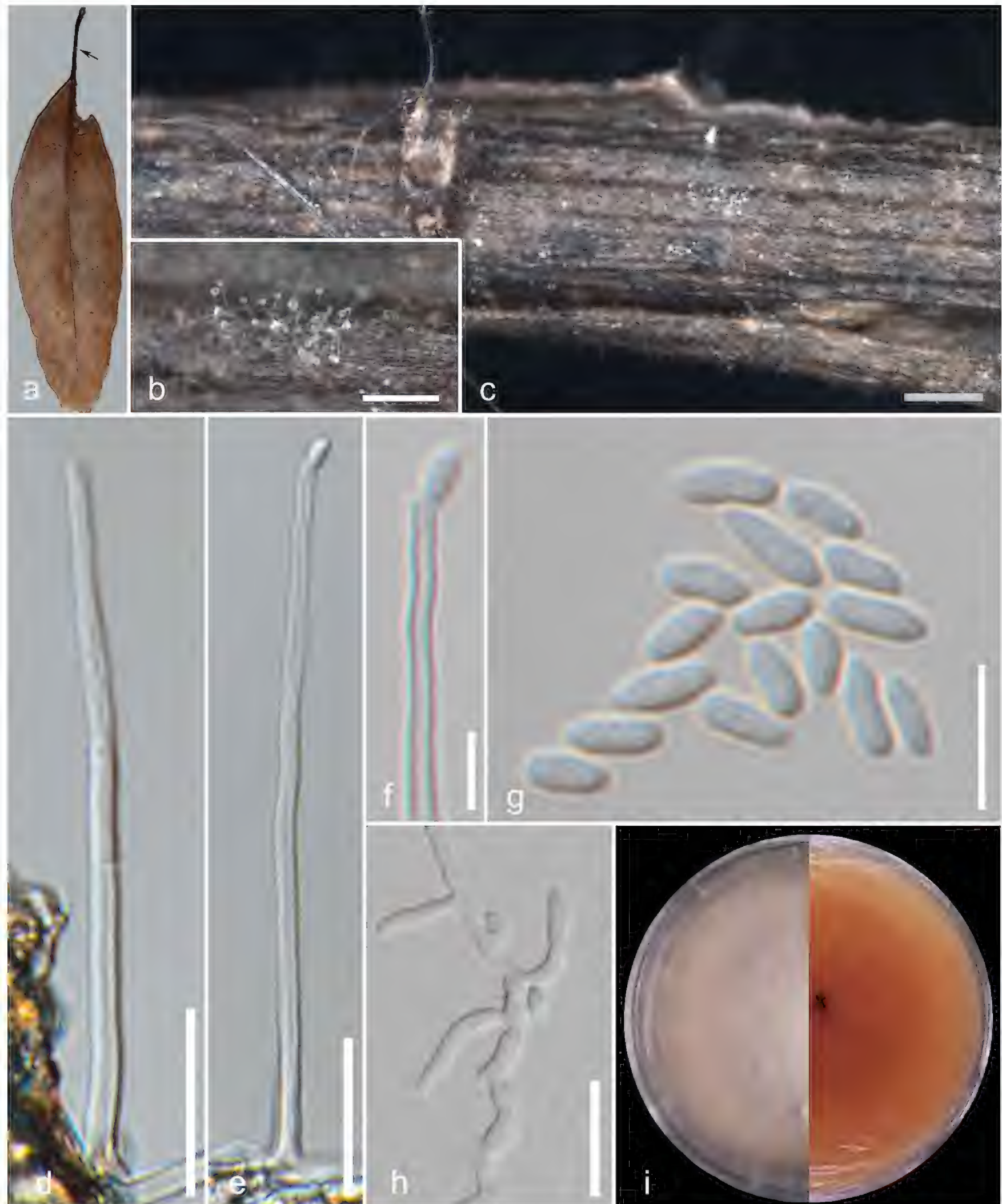


Figure 3. *Ochronectria thailandica* (MFLU 24–0383) **a** host (arrow points to the colonies on the petiole) **b**, **c** a closer view of the colonies on the host substrate **d–f** conidiophores with attached conidia **g** conidia **h** germinated spore **i** front and the reverse view of the culture on MEA media. Scale bars: 200 μ m (**b**); 500 μ m (**c**); 20 μ m (**d–e**, **h**); 5 μ m (**f**); 10 μ m (**g**).

combined in Fig. 4, using the BI tree as the base. The IQ-TREE web server's default settings were used for the maximum likelihood tree. At the same time, Tracer v1.7.2 was employed to assess the run quality of the BI tree, ensuring effective sampling size (ESS) values for all parameters. The alignment contained 1,096 unique sites (ITS: 262; LSU: 145; *rpb2*: 419; *tef1*– α : 270). Although the target species, *Ochronectria thailandica*, only had ITS and LSU sequences, all four loci were used in the multi-loci phylogenetic analysis, resulting in a stable tree.

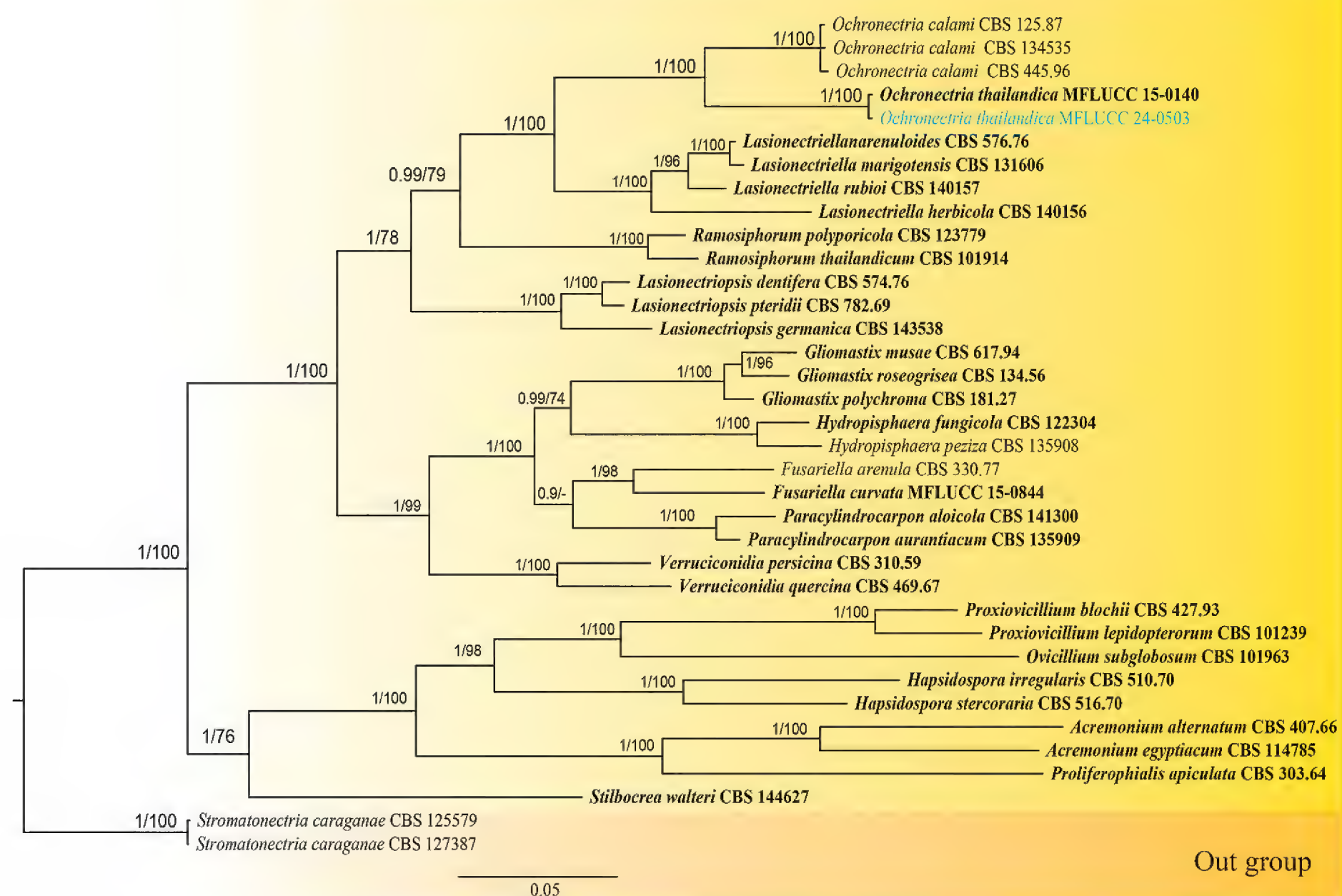


Figure 4. Bayesian inference phylogenetic tree generated from ITS, LSU, *rpb2*, and *tef1-α* sequence data for selected taxa from Bionectriaceae. The tree is rooted with two taxa from Stromatonectriaceae (CBS 125579 and CBS 127387). The newly generated sequence is in blue. Holotype and ex-type strains are in bold text. BYPP ≥ 0.95 and MLBS values $\geq 70\%$ are shown at the nodes.

Notes. *Ochronectria thailandica* was introduced by Li et al. (2016) in Chiang Rai Province, Thailand, from unidentified wood in the water. Our collection from Doi Inthanon includes an isolate found on the petiole of a fallen broadleaf species, which is an asexual morph. In the multi-locus phylogenetic analysis, this isolate clusters with the ex-type strain of *Ochronectria thailandica* (MFLUCC 15-0140) with BYPP of 1 and MLBS of 100% support. Based on base pair comparisons, the ITS region is identical (99%), with two gaps, and the LSU region is also identical (100%), with no gaps. This confirms that our isolate shares the same identity as the type strain of *Ochronectria thailandica*. As a result, we introduce the asexual morph of *O. thailandica* here, supported by graphical illustrations and morpho-phylogenetic evidence. Previously, *Ochronectria* was recognised as a genus with only a sexual morph, but our discovery establishes its holomorphic nature. Thus, we amend the genus description to include the asexual morph.

Discussion

This study introduces the new genus *Muciflexus* with *Muciflexus inthanonensis* sp. nov. and the asexual morph of *Ochronectria thailandica* based on a polyphasic approach, discovered during our ongoing exploration of fungal diversity in Doi Inthanon National Park.

The phylogeny of Niessliaceae remains unresolved (Hou et al. 2023), comprising several polyphyletic genera, such as *Niesslia*, *Cylindromonium*, and *Trichonectria*. The discovery of *Muciflexus* reveals a previously unknown lineage within Niessliaceae. Identifying new lineages like this is crucial for resolving unresolved taxonomic placements, as they may provide missing information needed for clarity. Previously, *Ochronectria* was known only for its sexual morph. The discovery of the asexual morph of *Ochronectria thailandica* extends our understanding of the genus by confirming its holomorphic nature. As a result, the genus description is amended to include the asexual morph characteristics. According to Species Fungorum (2025), *Ochronectria* includes three accepted species: *O. thailandica*, *O. calami*, and *O. courtecuissei*, which have been reported in Asia (Indonesia and Thailand) and Europe (France) across both terrestrial and freshwater environments (Rossman et al. 1999; Lechat 2010; Li et al. 2016). The sexual morph of *O. thailandica* was previously identified on decaying wood in freshwater habitats; in contrast, our study reports the asexual morph on a decaying dicotyledon leaf petiole in a terrestrial habitat (Li et al. 2016). This highlights the ecological versatility of *O. thailandica*, suggesting it can inhabit both freshwater and terrestrial environments. Consequently, *O. calami* and *O. courtecuissei* may also possess this habitat adaptability.

These findings provide deeper insight into the leaf litter fungal diversity of northern Thailand, confirming its potential to harbour more undiscovered species. The fungi were found in saprobic life forms, highlighting their role in nutrient recycling within the forest ecosystem. Additionally, revealing the versatile nature of *Ochronectria thailandica* to thrive in both aquatic and terrestrial environments further confirms the contribution of fungi to nutrient cycling in both ecosystems.

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Additional information

Conflict of interest

The authors have declared that no competing interests exist.

Ethical statement

No ethical statement was reported.

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Author contributions

Veenavee S. Hittanadurage Silva: Collecting, conceptualisation, original draft writing, and design of the study. Ruvishika S. Jayawardena: Supervision, guiding, reviewing, and editing. Rekhani H. Perera: Guiding, reviewing, and editing. Qirui Li: Guiding, reviewing, and editing. Kevin D. Hyde: Guiding, reviewing, and editing.

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Data availability

All of the data that support the findings of this study are available in the main text or Supplementary Information.

References

- Bucher VVC, Hyde KD, Pointing SB, Reddy CA (2004) Production of wood decay enzymes, mass loss and lignin solubilization in wood by marine ascomycetes and their anamorphs. *Fungal Diversity* 15: 14. <https://www.fungaldiversity.org/fdp/sfdp/15-1.pdf>
- Capella-Gutiérrez S, Silla-Martínez JM, Gabaldón T (2009) trimAl: A tool for automated alignment trimming in large-scale phylogenetic analyses. *Bioinformatics* (Oxford, England) 25: 1972–1973. <https://doi.org/10.1093/bioinformatics/btp348>
- Chomnunti P, Hongsanan S, Aguirre-Hudson B, Tian Q, Persoh D, Dhami M, Xu J, Liu X, Stadler M, Hyde K, Alias A (2014) The sooty moulds. *Fungal Diversity* 66: 1–36. <https://doi.org/10.1007/s13225-014-0278-5>
- Crous P, Wingfield M, Lombard L, Roets F, Swart W, Alvarado P, Carnegie A, Moreno G, Luangsa-Ard J, Thangavel R, Alexandrova A, Baseia I, Bellanger J-M, Bessette A, Bessette A, De la Peña Lastra S, García D, Gené J, Pham THG, Groenewald J (2019) Fungal Planet description sheets: 951–1041. *Persoonia - Molecular Phylogeny and Evolution of Fungi* 43: 223–425. <https://doi.org/10.3767/persoonia.2019.43.06>
- Crous PW, Carnegie AJ, Wingfield MJ, Sharma R, Mughini G, Noordeloos ME, Santini A, Shouche YS, Bezerra JDP, Dima B, Guarnaccia V, Imrefi I, Jurjević Ž, Knapp DG, Kovács GM, Magistà D, Perrone G, Rämä T, Rebriev YA, Shivas RG, Singh SM, Souza-Motta CM, Thangavel R, Adhasure NN, Alexandrova AV, Alfenas AC, Alfenas RF, Alvarado P, Alves AL, Andrade DA, Andrade JP, Barbosa RN, Barili A, Barnes CW, Baseia IG, Bellanger J-M, Berlanas C, Bessette AE, Bessette AR, Biketova AY, Bomfim FS, Brandrud TE, Bransgrove K, Brito ACQ, Cano-Lira JF, Cantillo T, Cavalcanti AD, Cheewangkoon R, Chikowski RS, Conforto C, Cordeiro TRL, Craine JD, Cruz R, Damm U, de Oliveira RJV, de Souza JT, de Souza HG, Dearnaley JDW, Dimitrov RA, Dovana F, Erhard A, Esteve-Raventós F, Félix CR, Ferisin G, Fernandes RA, Ferreira RJ, Ferro LO, Figueiredo CN, Frank JL, Freire KTLS, García D, Gené J, Gêsiorska A, Gibertoni TB, Gondra RAG, Gouliamova DE, Gramaje D, Guard F, Gusmão LFP, Haitook S, Hirooka Y, Houbaken J, Hubka V, Inamdar A, Iturriaga T, Iturrieta-González I, Jadan M, Jiang N, Justo A, Kachalkin AV, Kapitonov VI, Karadelev M, Karakehian J, Kasuya T, Kautmanová I, Kruse J, Kušan I, Kuznetsova TA, Landell MF, Larsson K-H, Lee HB, Lima DX, Lira

- CRS, Machado AR, Madrid H, Magalhães OMC, Majerova H, Malysheva EF, Mapper-son RR, Marbach PAS, Martín MP, Martín-Sanz A, Matočec N, McTaggart AR, Mello JF, Melo RFR, Mešić A, Michereff SJ, Miller AN, Minoshima A, Molinero-Ruiz L, Morozova OV, Mosoh D, Nabe M, Naik R, Nara K, Nascimento SS, Neves RP, Olariaga I, Oliveira RL, Oliveira TGL, Ono T, Ordoñez ME, Ottoni A de M, Paiva LM, Pancorbo F, Pant B, Pawłowska J, Peterson SW, Raudabaugh DB, Rodríguez-Andrade E, Rubio E, Rusevska K, Santiago ALCMA, Santos ACS, Santos C, Sazanov NA, Shah S, Sharma J, Silva BDB, Siquier JL, Sonawane MS, Stchigel AM, Svetasheva T, Tamakeaw N, Telleria MT, Tiago PV, Tian CM, Tkalčec Z, Tomashevskaya MA, Truong HH, Vecherskii MV, Visagie CM, Vizzini A, Yilmaz N, Zmitrovich IV, Zvyagina EA, Boekhout T, Kehlet T, Læssøe T, Groenewald JZ (2019a) Fungal Planet description sheets: 868–950. *Persoonia. Molecular Phylogeny and Evolution of Fungi* 42: 291–473. <https://doi.org/10.3767/persoonia.2019.42.11>
- Crous PW, Wingfield MJ, Lombard L, Roets F, Swart WJ, Roets F, Swart WJ, Alvarado P, Carnegie AJ, Moreno G, Luangsa-Ard J, Thangavel R, Alexandrova AV, Baseia IG, Bellanger J-M, Bessette AE, Bessette AR, Delapeña-Lastra S, García D, Gené J, Pham THG, Heykoop M, Malysheva E, Malysheva V, Martín MP, Morozova OV, Noisripoom W, Overton BE, Rea AE, Sewall BJ, Smith ME, Smyth CW, Tasanathai K, Visagie CM, Adamčík S, Alves A, Andrade JP, Aninat MJ, Araújo RVB, Bordallo JJ, Bouffleur T, Bar-oncelli R, Barreto RW, Bolin J, Cabero J, Caboñ M, Cafà G, Caffot MLH, Cai L, Carlavilla JR, Chávez R, Decastro RRL, Delgat L, Deschuyteneer D, Dios MM, Domínguez LS, Evans HC, Eyssartier G, Ferreira BW, Figueiredo CN, Liu F, Fournier J, Galli-Terasawa LV, Gil-Durán C, Glienke C, Gonçalves MFM, Gryta H, Guarro J, Himaman W, Hywel-Jones N, Iturrieta-González I, Ivanushkina NE, Jargeat P, Khalid AN, Khan J, Kiran M, Kiss L, Kochkina GA, Kolařík M, Kubátová A, Lodge DJ, Loizides M, Luque D, Manjón JL, Marbach PAS, Massolajr NS, Mata M, Miller AN, Mongkolsamrit S, Moreau P-A, Morte A, Mujic A, Navarro-Ródenas A, Németh MZ, Nóbrega TF, Nováková A, Olariaga I, Ozerskaya SM, Palma MA, Petters-Vandresen DAL, Piontelli E, Popov ES, Rodríguez A, Requejo Ó, Rodrigues ACM, Rong IH, Roux J, Seifert KA, Silva BDB, Sklenář F, Smith JA, Sousa JO, Souza HG, Desouza JT, Švec K, Tanchaud P, Tanney JB, Terasawa F, Thanakitpipattana D, Torres-Garcia D, Vaca I, Vaghefi N, Vaniperen AL, Vasilenko OV, Verbeken A, Yilmaz N, Zamora JC, Zapata M, Jurjevi Ž, Groenewald JZ (2019b) Fungal Planet description sheets: 951–1041. *Persoonia* 43: 223–425. <https://doi.org/10.3767/persoonia.2019.43.06>
- Crous PW, Cowan DA, Maggs-Kolling G, Yilmaz N, Larsson E, Angelini C, Brandrud TE, Dearnaley JDW, Dima B, Dovana F, Fechner N, García D, Gené J, Halling RE, Houbraken J, Leonard P, Luangsa-ard JJ, Noisripoom W, Rea-Ireland AE, Ševčíková H, Smyth CW, Vizzini A, Adam JD, Adams GC, Alexandrova AV, Alizadeh A, Álvarez Duarte E, Andjic V, Antonín V, Arenas F, Assabgui R, Ballarà J, Banwell A, Berraf-Tebbal A, Bhatt VK, Bonito G, Botha W, Burgess TI, Caboñ M, Calvert J, Carvalhais LC, Courtecuisse R, Cullington P, Davoodian N, Decock CA, Dimitrov R, Di Piazza S, Drenth A, Dumez S, Eichmeier A, Etayo J, Fernández I, Fiard J-P, Fournier J, Fuentes-Aponte S, Ghanbary MAT, Ghorbani G, Giraldo A, Glushakova AM, Gouliamova DE, Guarro J, Halleen F, Hampe F, Hernández-Restrepo M, Iturrieta-González I, Jeppson M, Kachalkin AV, Karimi O, Khalid AN, Khonsanit A, Kim JI, Kim K, Kiran M, Krisai-Greilhuber I, Kučera V, Kušan I, Langenhoven SD, Lebel T, Lebeuf R, Liimatainen K, Linde C, Lindner DL, Lombard L, Mahamedi AE, Matočec N, Maxwell A, May TW, McTaggart AR, Meijer M, Mešić A, Mileto AJ, Miller AN, Molia A, Mongkolsamrit S, Muñoz Cortés C, Muñoz-Mohedano J, Morte A, Morozova OV, Mostert L, Mostowfizadeh-Ghalamfarsa R, Nagy LG, Navarro-

- Ródenas A, Örstadius L, Overton BE, Papp V, Para R, Peintner U, Pham THG, Pordel A, Pošta A, Rodríguez A, Romberg M, Sandoval-Denis M, Seifert KA, Semwal KC, Sewall BJ, Shivas RG, Slovák M, Smith K, Spetik M, Spies CFJ, Syme K, Tasanathai K, Thorn RG, Tkalčec Z, Tomashevskaya MA, Torres-Garcia D, Ullah Z, Visagie CM, Voitek A, Winton LM, Groenewald JZ (2020) Fungal Planet description sheets: 1112–1181. *Persoonia* 45(1): 251–409. <https://doi.org/10.3767/persoonia.2020.45.10>
- Darriba D, Taboada GL, Doallo R, Posada D (2012) jModelTest 2: More models, new heuristics and parallel computing. *Nature Methods* 9: 772–772. <https://doi.org/10.1038/nmeth.2109>
- Du E, Terrer C, McNulty SG, Jackson RB (2024) Nutrient limitation in global forests: current status and future trends. In: *Future Forests*. Elsevier, 65–74. <https://doi.org/10.1016/B978-0-323-90430-8.00014-9>
- Flakus A, Etayo J, Miadlikowska J, Lutzoni F, Kukwa M, Matura N, Rodriguez-Flakus P (2019) Biodiversity assessment of ascomycetes inhabiting Lobariella lichens in Andean cloud forests led to one new family, three new genera and 13 new species of lichenicolous fungi. *Plant and Fungal Systematics* 64: 283–344. <https://doi.org/10.2478/pfs-2019-0022>
- Guindon S, Gascuel O (2003) A Simple, Fast, and Accurate Algorithm to Estimate Large Phylogenies by Maximum Likelihood. *Systematic Biology* 52: 696–704. <https://doi.org/10.1080/10635150390235520>
- Hou LW, Giraldo A, Groenewald JZ, Rämä T, Summerbell RC, Huang GZ, Cai L, Crous PW (2023) Redisposition of acremonium-like fungi in Hypocreales. *Studies in Mycology* 105: 23–203. <https://doi.org/10.3114/sim.2023.105.02>
- Huelsenbeck JP, Ronquist F (2001) MRBAYES: Bayesian inference of phylogenetic trees. *Bioinformatics* 17: 754–755. <https://doi.org/10.1093/bioinformatics/17.8.754>
- Hyde KD, Norphanphoun C, Chen J, Dissanayake AJ, Doilom M, Hongsan S, Jayawardena RS, Jeewon R, Perera RH, Thongbai B, Wanasinghe DN, Wisitrassameewong K, Tibpromma S, Stadler M (2018) Thailand’s amazing diversity: Up to 96% of fungi in northern Thailand may be novel. *Fungal Diversity* 93: 215–239. <https://doi.org/10.1007/s13225-018-0415-7>
- Hyde KD, Xu J, Rapior S, Jeewon R, Lumyong S, Niego AGT, Abeywickrama PD, Aluthmuhandiram JVS, Brahamanage RS, Brooks S, Chaiyasen A, Chethana KWT, Chomnunti P, Chepkirui C, Chuankid B, De Silva NI, Doilom M, Faulds C, Gentekaki E, Gopalan V, Kakumyan P, Harishchandra D, Hemachandran H, Hongsan S, Karunarathna A, Karunarathna SC, Khan S, Kumla J, Jayawardena RS, Liu J-K, Liu N, Luangharn T, Macabeo APG, Marasinghe DS, Meeks D, Mortimer PE, Mueller P, Nadir S, Nataraja KN, Nontachaiyapoom S, O’Brien M, Penkhruue W, Phukhamsakda C, Ramanan US, Rathnayaka AR, Sadaba RB, Sandargo B, Samarakoon BC, Tennakoon DS, Siva R, Sriprom W, Suryanarayanan TS, Sujarit K, Suwannarach N, Suwunwong T, Thongbai B, Thongklang N, Wei D, Wijesinghe SN, Winiski J, Yan J, Yasanthika E, Stadler M (2019) The amazing potential of fungi: 50 ways we can exploit fungi industrially. *Fungal Diversity* 97: 1–136. <https://doi.org/10.1007/s13225-019-00430-9>
- Hyde KD, Norphanphoun C, Maharachchikumbura S, Bhat DJ, Jones EBG, Bundhun D, Chen YJ, Bao D-F, Boonmee S, Calabon M, Chaiwan N, Chethana KWT, Dai D-Q, Dayarathne MC, Devadatha B, Dissanayake AJ, Dissanayake LS, Doilom M, Dong W, Zhang S, Fan X, Goonasekara ID, Hongsan S, Huang SK, Jayawardena RS, Jeewon R, Karunarathna A, Konta S, Kumar V, Lin CG, Liu J-K, Liu NG, Luangsa-ard J, Lumyong S, Luo ZL, Marasinghe DS, McKenzie EHC, Niego AGT, Niranjana M, Perera RH, Phukhamsakda C, Rathnayaka A, Samarakoon MC, Samarakoon S, Sarma VV,

- Senanayake IC, Shang QJ, Stadler M, Tibpromma S, Wanasinghe DN, Wei D, Wijayawardene NN, Xiao YP, Yang J, Zeng XY, Xiang MM (2020) Refined families of Sordariomycetes. *Mycosphere* 11: 1058. <https://doi.org/10.5943/mycosphere/11/1/7>
- Hyde KD, Noorabadi MT, Thiyagaraja V, He MQ, Johnston PR, Wijesinghe SN, Armand A, Biketova AY, Chethana KWT, Erdoğan M, Ge ZW, Groenewald JZ, Hongsan S, Kušan I, Leontyev DV, Li DW, Lin CG, Liu NG, Maharachchikumbura SSN, Matočec N, May TW, McKenzie EHC, Mešić A, Perera RH, Phukhamsakda C, Piątek M, Samarakoon MC, Selcuk F, Senanayake IC, Tanney JB, Tian Q, Vizzini A, Wanasinghe DN, Wannasawang N, Wijayawardene NN, Zhao RL, Abdel-Wahab MA, Abdollahzadeh J, Abeywickrama PD, Abhinav, Absalan S, Acharya K, Afshari N, Afshan NS, Afzalinia S, Ahmadpour SA, Akulov O, Alizadeh A, Alizadeh M, Al-Sadi AM, Alves A, Alves VCS, Alves-Silva G, Antonín V, Aouali S, Aptroot A, Apurillo CCS, Arias RM, Asgari, B, Asghari R, Assis DMA, Assyov B, Atienza V, Aumentado HDR, Avasthi S, Azevedo E, Bakhshi M, Bao DF, Baral HO, Barata M, Barbosa KD, Barbosa RN, Barbosa FR, Baroncelli R, Barreto GG, Baschien C, Bennett RM, Bera I, Bezerra JDP, Bhunjun CS, Bianchinotti MV, Błaszczowski J, Boekhout T, Bonito GM, Boonmee S, Boonyuen N, Bortnikov FM, Bregant C, Bundhun D, Burgaud G, Buyck B, Caeiro MF, Cabarroi-Hernández M, Cai M Feng, Cai L, Calabon MS, Calaça FJS, Callalli M, Câmara MPS, Cano-Lira J, Cao B, Carlavilla JR, Carvalho A, Carvalho TG, Castañeda-Ruiz RF, Catania MDV, Cazabonne J, Cedeño-Sanchez M, Chaharmiri-Dokhaharani S, Chaiwan N, Chakraborty N, Cheewankoon R, Chen C, Chen J, Chen Q, Chen YP, Chinaglia S, Coelho-Nascimento CC, Coleine C, CostaRezende DH, Cortés-Pérez A, Crouch, JA, Crous PW, Cruz RHSF, Czachura P, Damm U, Darmostuk V, Daroodi Z, Das K, Das K, Davoodian N, Davydov EA, da Silva GA, da Silva IR, da Silva RMF, da Silva Santos AC, Dai DQ, Dai YC, de Groot Michiel D, De Kesel A, De Lange R, de Medeiros EV, de Souza CFA, de Souza FA, dela Cruz TEE, Decock C, Delgado G, Denchev CM, Denchev TT, Deng YL, Dentinger BTM, Devadatha B, Dianese JC, Dima B, Doilom M, Dissanayake AJ, Dissanayake DMLS, Dissanayake LS, Diniz AG, Dolatabadi S, Dong JH, Dong W, Dong ZY, Drechsler-Santos ER, Druzhinina IS, Du TY, Dubey MK, Dutta AK, Elliott TF, Elshahed MS, Egidi E, Eisvand P, Fan L, Fan X, Fan XL, Fedosova AG, Ferro LO, Fiuza PO, Flakus A, W. Fonseca EO, Fryar SC, Gabaldón T, Gajanayake AJ, Gannibal PB, Gao F, GarcíaSánchez D, García-Sandoval R, Garrido-Benavent I, Garzoli L, Gasca-Pineda J, Gautam AK, Gené J, Ghobad-Nejhad M, Ghosh A, Giachini AJ, Gibertoni TB, Gentekaki E, Gmoshinskiy VI, GóesNeto A, Gomdola D, Gorjón SP, Goto BT, Granados-Montero MM, Griffith GW, Groenewald M, Grossart H-P, Gu ZR, Gueidan C, Gunarathne A, Gunaseelan S, Guo SL, Gusmão LFP, Gutierrez AC, Guzmán-Dávalos L, Haelewaters D, Haituk H, Halling RE, He SC, Heredia G, HernándezRestrepo M, Hosoya T, Hoog SD, Horak E, Hou CL, Houbraken J, Htet ZH, Huang SK, Huang WJ, Hurdeal VG, Hustad VP, Inácio CA, Janik P, Jayalal RGU, Jayasiri SC, Jayawardena RS, Jeewon R, Jerônimo GH, Jin J, Jones EBG, Joshi Y, Jurjević Ž, Justo A, Kakishima M, Kaliyaperumal M, Kang GP, Kang JC, Karimi O, Karunarathna SC, Karpov SA, Kezo K, Khalid AN, Khan MK, Khuna S, Khyaju S, Kirchmair M, Klawonn I, Kraisitudomsook N, Kukwa M, Kularathnage ND, Kumar S, Lachance MA, Lado C, Latha KPD, Lee HB, Leonardi M, Lestari AS, Li C, Li H, Li J, Li Q, Li Y, Li YC, Li YX, Liao CF, Lima JLR, Lima JMS, Lima NB, Lin L, Linaldeddu BT, Linn MM, Liu F, Liu JK, Liu JW, Liu S, Liu SL, Liu XF, Liu XY, Longcore JE, Luangharn T, Luangsa-ard JJ, Lu L, Lu YZ, Lumbsch HT, Luo L, Luo M, Luo ZL, Ma J, Madagammana AD, Madhushan A, Madrid H, Magurno F, Magyar D, Mahadevakumar S, Malosso E, Malysch JM, Mamarabadi M, Manawasinghe IS, Manfrino RG, Manimohan P, Mao N, Mapook A, Marchese P, Marasinghe DS, Mardones M, Marin-Felix Y, Masigol H, Mehrabi M, MehrabiKoushki M, Meiras-Ottoni A de, Melo RFR, Mendes-Alvarenga RL, Mendieta S, Meng QF, Menkis A,

- Menolli Jr N, Mikšík M, Miller SL, Moncada B, Moncalvo JM, Monteiro JS, Monteiro M, Mora-Montes HM, Moroz EL, Moura JC, Muhammad U, Mukhopadhyay S, Nagy GL, Najam ul Sehar A, Najafiniya M, Nanayakkara CM, Naseer A, Nascimento ECR, Nascimento SS, Neuhauser S, Neves MA, Niazi AR, Nie Yong, Nilsson RH, Nogueira PTS, Novozhilov YK, Noordeloos M, Norphanphoun C, Nuñez Otaño N, O'Donnell RP, Oehl F, Oliveira JA, Oliveira Junior I, Oliveira NVL, Oliveira PHF, Orihara T, Oset M, Pang KL, Papp V, Pathirana LS, Peintner U, Pem D, Pereira OL, Pérez-Moreno J, Pérez-Ortega S, Péter G, Pires-Zotarelli CLA, Phonemany M, Phongeun S, Pošta A, Prazeres JFSA, Quan Y, Quandt CA, Queiroz MB, Radek R, Rahnama K, Raj KNA, Rajeshkumar KC, Rajwar Soumyadeep, Ralaiveloarisoa AB, Rämä T, Ramírez-Cruz V, Rambold G, Rathnayaka AR, Raza M, Ren GC, Rinaldi AC, Rivas-Ferreiro M, Robledo GL, Ronikier A, Rossi W, Rusevska K, Ryberg M, Safi A, Salimi F, Salvador-Montoya CA, Samant B, Samaradiwakara NP, Sánchez-Castro I, Sandoval-Denis M, Santiago ALCMA, Santos ACDS, Santos LA dos, Sarma VV, Sarwar S, Savchenko A, Savchenko K, Saxena RK, Schoutteten N, Selbmann L, Ševčíková H, Sharma A, Shen HW, Shen YM, Shu YX, Silva HF, Silva-Filho AGS, Silva VSH, Simmons DR, Singh R, Sir EB, Sohrabi M, Souza FA, Souza-Motta CM, Sriindrasutdhi V, Sruthi OP, Stadler M, Stemler J, Stephenson SL, Stoyneva-Gaertner MP, Strasser JFH, Stryjak-Bogacka M, Su H, Sun YR, Svantesson S, Sysouphanthong P, Takamatsu S, Tan TH, Tanaka K, Tang C, Tang X, Taylor JE, Taylor PWJ, Tennakoon DS, Thakshila SAD, Thambugala KM, Thamodini GK, Thilanga D, Thines M, Tiago PV, Tian XG, Tian WH, Tibpromma S, Tkalčec Z, Tokarev YS, Tomšovský M, Torruella G, Tsurykau A, Udayanga D, Ulukapı M, Untereiner WA, Usman M, Uzunov BA, Vadthanarat S, Valenzuela R, Van den Wyngaert S, Van Vooren N, Velez P, Verma RK, Vieira LC, Vieira WAS, Vinzelj JM, Tang AMC, Walker A, Walker AK, Wang QM, Wang Y, Wang XY, Wang ZY, Wannathes N, Wartchow F, Weerakoon G, Wei DP, Wei X, White JF, Wijesundara DSA, Wisitrassameewong K, Worobiec G, Wu HX, Wu N, Xiong YR, Xu B, Xu JP, Xu R, Xu RF, Xu RJ, Yadav S, Yakovchenko LS, Yang HD, Yang X, Yang YH, Yang Y, Yang YY, Yoshioka R, Youssef Noha H, Yu FM, Yu ZF, Yuan LL, Yuan Q, Zabin DA, Zamora JC, Zapata CV, Zare R, Zeng M, Zeng XY, Zhang JF, Zhang JY, Zhang S, Zhang XC, Zhao CL, Zhao H, Zhao Q, Zhao H, Zhao HJ, Zhou HM, Zhu XY, Zmitrovich IV, Zucconi L, Zvyagina E (2024) The 2024 Outline of Fungi and fungus-like taxa. *Mycosphere* 15(1): 5146–6239. <https://doi.org/10.5943/mycosphere/15/1/25>
- Index Fungorum (2025) Index Fungorum. <https://www.indexfungorum.org/names/Names.asp>
- Katoh K, Rozewicki J, Yamada KD (2019) MAFFT online service: Multiple sequence alignment, interactive sequence choice and visualization. *Briefings in Bioinformatics* 20: 1160–1166. <https://doi.org/10.1093/bib/bbx108>
- Kimura M (1980) A simple method for estimating evolutionary rates of base substitutions through comparative studies of nucleotide sequences. *Journal of Molecular Evolution* 16: 111–120. <https://doi.org/10.1007/BF01731581>
- Kjøller A, Struwe S, Kjøller A (1982) Microfungi in Ecosystems: Fungal Occurrence and Activity in Litter and Soil. *Oikos* 39: 391. <https://doi.org/10.2307/3544690>
- Kumar S, Stecher G, Suleski M, Sanderford M, Sharma S, Tamura K (2024) MEGA12: Molecular Evolutionary Genetic Analysis Version 12 for Adaptive and Green Computing. *Molecular Biology and Evolution* 41. [March 11, 2025] <https://doi.org/10.1093/molbev/msae263>
- Kuraku S, Zmasek CM, Nishimura O, Katoh K (2013) aLeaves facilitates on-demand exploration of metazoan gene family trees on MAFFT sequence alignment server with enhanced interactivity. *Nucleic Acids Research* 41: W22–W28. <https://doi.org/10.1093/nar/gkt389>

- Lechat C (2010) *Ochronectria courtecuissei* sp. nov. Bulletin Trimestriel de la Societe Mycologique de France 126: 97. http://ascofrance.com/uploads/forum_file/BSMF-2010-v126-p97-0001.pdf
- Li GJ, Hyde KD, Zhao RL, Hongsanan S, Abdel-Aziz FA, Abdel-Wahab MA, Alvarado P, Alves-Silva G, Ammirati JF, Ariyawansa HA, Baghela A, Bahkali AH, Beug M, Bhat DJ, Bojantchev D, Boonpratuang T, Bulgakov TS, Camporesi E, Boro MC, Ceska O, Chakraborty D, Chen JJ, Chethana KWT, Chomnunti P, Consiglio G, Cui BK, Dai DQ, Dai YC, Daranagama DA, Das K, Dayarathne MC, De Crop E, De Oliveira RJV, de Souza CAF, de Souza JI, Dentinger BTM, Dissanayake AJ, Doilom M, Drechsler-Santos ER, Ghobad-Nejhad M, Gilmore SP, Góes-Neto A, Gorczak M, Haitjema CH, Hapuarachchi KK, Hashimoto A, He MQ, Henske JK, Hirayama K, Iribarren MJ, Jayasiri SC, Jayawardena RS, Jeon SJ, Jerônimo GH, Jesus AL, Jones EBG, Kang JC, Karunarathna SC, Kirk PM, Konta S, Kuhnert E, Langer E, Lee HS, Lee HB, Li WJ, Li XH, Liimatainen K, Lima DX, Lin CG, Liu JK, Liu XZ, Liu ZY, Luangsa-ard JJ, Lücking R, Lumbsch HT, Lumyong S, Leaño EM, Marano AV, Matsumura M, McKenzie EHC, Mongkolsamrit S, Mortimer PE, Nguyen TTT, Niskanen T, Norphanphoun C, O'Malley MA, Parnmen S, Pawłowska J, Perera RH, Phookamsak R, Phukhamsakda C, Pires-Zottarelli CLA, Raspé O, Reck MA, Rocha SCO, de Santiago ALCMA, Senanayake IC, Setti L, Shang QJ, Singh SK, Sir EB, Solomon KV, Song J, Srikukulchai P, Stadler M, Suetrong S, Takahashi H, Takahashi T, Tanaka K, Tang LP, Thambugala KM, Thanakitpipattana D, Theodorou MK, Thongbai B, Thummarukcharoen T, Tian Q, Tibpromma S, Verbeken A, Vizzini A, Vlasák J, Voigt K, Wanasinghe DN, Wang Y, Weerakoon G, Wen HA, Wen TC, Wijayawardene NN, Wongkanoun S, Wrzosek M, Xiao YP, Xu JC, Yan JY, Yang J, Da Yang S, Hu Y, Zhang JF, Zhao J, Zhou LW, Peršoh D, Phillips AJL, Maharachchikumbura SSN (2016) Fungal diversity notes 253–366: Taxonomic and phylogenetic contributions to fungal taxa. Fungal Diversity 78: 1–237. <https://doi.org/10.1007/s13225-016-0366-9>
- Likens GE (2013) Biogeochemistry of a Forested Ecosystem. Springer Science & Business Media, 234 pp. <https://doi.org/10.1007/978-1-4614-7810-2>
- Lombard L, Van der Merwe NA, Groenewald JZ, Crous PW (2015) Generic concepts in Nectriaceae. Studies in Mycology 80: 189–245. <https://doi.org/10.1016/j.simyco.2014.12.002>
- Louangphan J, Perera R, Farias A (2024) A new addition to Kirschsteinioteliaceae: *Kirschsteiniotelia Chiangmaiensis* sp. nov. from Northern Thailand. Phytotaxa 634: 49–62. <https://doi.org/10.11646/phytotaxa.634.1.4>
- Maharachchikumbura SSN, Chen Y, Ariyawansa HA, Hyde KD, Haelewaters D, Perera RH, Samarakoon MC, Wanasinghe DN, Bustamante DE, Liu J-K, Lawrence DP, Cheewangkoon R, Stadler M (2021) Integrative approaches for species delimitation in Ascomycota. Fungal Diversity 109: 155–179. <https://doi.org/10.1007/s13225-021-00486-6>
- Niego AG, Lambert C, Mortimer P, Thongklang N, Rapior S, Grosse M, Schrey H, Charria-Girón E, Walker A, Hyde K, Stadler M (2023) The contribution of fungi to the global economy. Fungal Diversity 121: 95–137. <https://doi.org/10.1007/s13225-023-00520-9>
- Osono T (2007) Ecology of ligninolytic fungi associated with leaf litter decomposition. Ecological Research 22: 955–974. <https://doi.org/10.1007/s11284-007-0390-z>
- Perera RH, Hyde KD, Jones EBG, Maharachchikumbura SSN, Bundhun D, Camporesi E, Akulov A, Liu JK, Liu ZY (2023) Profile of Bionectriaceae, Calcarisporiaceae, Hypocreaceae, Nectriaceae, Tilachlidiaceae, Ijuhyaceae fam. nov., Stromatonectriaceae fam. nov. and Xanthonectriaceae fam. nov. Fungal Diversity 118: 95–271. <https://doi.org/10.1007/s13225-022-00512-1>

- Raja H, Miller A, Pearce C, Oberlies N (2017) Fungal Identification Using Molecular Tools: A Primer for the Natural Products Research Community. *Journal of Natural Products* 80. <https://doi.org/10.1021/acs.jnatprod.6b01085>
- Rambaut A (2012) FigTree v1. 4.0. <http://ac.uk/software/figtree>
- Rathnayaka AR, Tennakoon DS, Jones GEB, Wanasinghe DN, Bhat DJ, Priyashantha AKH, Stephenson SL, Tibpromma S, Karunarathna SC (2024) Significance of precise documentation of hosts and geospatial data of fungal collections, with an emphasis on plant-associated fungi. *New Zealand Journal of Botany* 31: 1–28. <https://doi.org/10.1080/0028825X.2024.2381734>
- Rehner S, Buckley E (2005) A *Beauveria* phylogeny inferred from nuclear ITS and EF1-?? sequences: Evidence for cryptic diversification and links to *Cordyceps* teleomorphs. *Mycologia* 97: 84–98. <https://doi.org/10.3852/mycologia.97.1.84>
- Rehner SA, Samuels GJ (1994) Taxonomy and phylogeny of *Gliocladium* analysed from nuclear large subunit ribosomal DNA sequences. *Mycological Research* 98: 625–634. [https://doi.org/10.1016/S0953-7562\(09\)80409-7](https://doi.org/10.1016/S0953-7562(09)80409-7)
- Ronquist F, Huelsenbeck JP (2003) MrBayes 3: Bayesian phylogenetic inference under mixed models. *Bioinformatics* 19: 1572–1574. <https://doi.org/10.1093/bioinformatics/btg180>
- Ronquist F, Teslenko M, Van Der Mark P, Ayres DL, Darling A, Höhna S, Larget B, Liu L, Suchard MA, Huelsenbeck JP (2012) MrBayes 3.2: efficient Bayesian phylogenetic inference and model choice across a large model space. *Systematic biology* 61: 539–542. <https://doi.org/10.1093/sysbio/sys029>
- Rossmann AY (1996) Morphological and molecular perspectives on systematics of the Hypocreales. *Mycologia* 88: 1–19. <https://doi.org/10.1080/00275514.1996.12026620>
- Rossmann AY, Samuels GJ, Rogerson CT, Lowen R (1999) Genera of Bionectriaceae, Hypocreaceae and Nectriaceae (Hypocreales, Ascomycetes). *Studies in Mycology* 42.
- Sayers EW, Cavanaugh M, Clark K, Ostell J, Pruitt KD, Karsch-Mizrachi I (2020) GenBank. *Nucleic Acids Research* 48: D84–D86. <https://doi.org/10.1093/nar/gkz956>
- Senanayake I, Rathnayaka A, Sandamali D, Calabon M, Gentekaki E, Lee H, Pem D, Dissanayake L, Wijesinghe S, Bundhun D, TTT N, Goonasekara I, Abeywickrama P, Jayawardena R, Wanasinghe D, Jeewon R, Bhat DJ, Mm X, Bhunjun CS, Hurdeal V (2020) Morphological approaches in studying fungi: Collection, examination, isolation, sporulation and preservation. *Mycosphere* 11: 2678–2754. <https://doi.org/10.5943/mycosphere/11/1/20>
- Silva H, Perera R, Farias A (2023) Addition to Pseudoplagiostomataceae: *Pseudoplagiostoma inthanonense* sp. nov. from Doi Inthanon National Park, Northern Thailand. *Phytotaxa* 625: 66–76. <https://doi.org/10.11646/phytotaxa.625.1.4>
- Species Fungorum (2025) Species Fungorum. <https://www.speciesfungorum.org/Names/Names.asp>
- Staden R, Judge DP, Bonfield JK (2003) Analyzing Sequences Using the Staden Package and EMBOSS. *Introduction to Bioinformatics: A Theoretical and Practical Approach*, 746 pp. https://doi.org/10.1007/978-1-59259-335-4_24
- Sun J, Yu S, Lu Y, Liu H, Liu X (2023) Proposal of a new family Pseudodiploösporeaceae fam. nov. (Hypocreales) based on phylogeny of *Diploöspora longispora* and *Paecilomyces penicillatus*. *Mycology* 14: 60–73. <https://doi.org/10.1080/21501203.2022.2143919>

- Sung G-H, Sung J-M, Hywel-Jones NL, Spatafora JW (2007) A multi-gene phylogeny of Clavicipitaceae (Ascomycota, Fungi): Identification of localized incongruence using a combinational bootstrap approach. *Molecular Phylogenetics and Evolution* 44: 1204–1223. <https://doi.org/10.1016/j.ympev.2007.03.011>
- Teejuntuk S, Sahunalu P, Sakurai K, Sungpalee W (2003) Forest Structure and Tree Species Diversity along an Altitudinal Gradient in Doi Inthanon National Park, Northern Thailand. *Tropics* 12: 85–102. <https://doi.org/10.3759/tropics.12.85>
- Thitla T, Kumla J, Khuna S, Lumyong S, Suwannarach N (2022) Species Diversity, Distribution, and Phylogeny of *Exophiala* with the Addition of Four New Species from Thailand. *Journal of Fungi (Basel, Switzerland)* 8: 766. <https://doi.org/10.3390/jof8080766>
- Trifinopoulos J, Nguyen L-T, von Haeseler A, Minh BQ (2016) W-IQ-TREE: a fast online phylogenetic tool for maximum likelihood analysis. *Nucleic Acids Research* 44: W232–W235. <https://doi.org/10.1093/nar/gkw256>
- Vaidya G, Lohman DJ, Meier R (2011) SequenceMatrix: Concatenation software for the fast assembly of multi-gene datasets with character set and codon information. *Cladistics* 27: 171–180. <https://doi.org/10.1111/j.1096-0031.2010.00329.x>
- Vilgalys R, Hester M (1990) Rapid genetic identification and mapping of enzymatically amplified ribosomal DNA from several *Cryptococcus* species. *Journal of Bacteriology* 172: 4238–4246. <https://doi.org/10.1128/jb.172.8.4238-4246.1990>
- White TJ, Bruns T, Lee S, Taylor J (1990) Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. *PCR protocols: a guide to methods and applications*, 482 pp. <https://doi.org/10.1016/B978-0-12-372180-8.50042-1>
- Wood TE, Lawrence D, Clark DA, Chazdon RL (2009) Rain forest nutrient cycling and productivity in response to large-scale litter manipulation. *Ecology* 90: 109–121. <https://doi.org/10.1890/07-1146.1>

Supplementary material 1

Niessliaceae and Bionectriaceae taxa table

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Data type: xlsx

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Supplementary material 2

Asexual morph comparison

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Data type: xlsx

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Supplementary material 3

Pairwise distance calculation of *Cylindromonium* and *Phialoseptomonium*

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Data type: xlsx

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